

### AMENDMENTS TO THE CLAIMS

1-84. Cancelled.

85. **(New)** A method for assessing the binding affinity between a candidate peptide and a target protein, said method comprising:

- obtaining a collection of sequence and binding strength data for at least one peptide of known affinity for said target protein;

- obtaining sequence data for said candidate peptide;

- determining a first affinity for the candidate peptide for said target protein using a first predictive method, wherein said determination is based, in part, upon the candidate peptide sequence data;

- determining a second affinity for the candidate peptide for said target protein using a second predictive method, wherein said determination is based, in part, upon the candidate peptide sequence data, said second predictive method further comprising the use of said collection of sequence and binding strength data, and wherein said second predictive method is different from the first predictive method;

- combining said first and second affinities; and

- evaluating the combined first and second affinities as a basis for assessing the binding affinity between the candidate peptide and the target protein.

86. **(New)** The method of Claim 85, wherein said target protein is a MHC class I protein and wherein said peptide comprises an epitope whereby said MHC class I protein binds to said peptide.

87. **(New)** The method of Claim 85, wherein the first and second affinities are scaled before they are combined, said scaling comprising a method selected from the group consisting of 1) linearly scaling each affinity so that it has a value between 1 and 0, 2) nonlinearly scaling each affinity so that it has a value between 1 and 0, and 3) scaling the affinity in a manner so a particular type of method can have a different weight, wherein a value is maintained between 1 and 0.

88. **(New)** The method of Claim 85, wherein the candidate peptide is generated by dividing the sequence data of a known protein into ninemer or tenmer fragments.

89. (New) The method of Claim 85, wherein said first predictive method and said second predictive method are selected from the group consisting of quadratic programming, linear programming, anchor scoring, and profile-based scoring.

90. (New) A method for evaluating the affinity of a candidate peptide for a target protein, said method comprising:

- obtaining for a plurality of known peptides, a collection of sequence and binding affinity information for said target protein;

- obtaining sequence data for the candidate peptide;

- predicting a first affinity for said candidate peptide for said target protein based, in part, upon the evaluation of the collection of sequence and binding affinity information for the plurality of known peptides;

- predicting a second affinity for said candidate peptide for said target protein based, in part, upon the evaluation of the collection of sequence and binding affinity information for the plurality of known peptides in a manner that differs from the first affinity prediction;

- normalizing said first affinity to generate a first vote;

- normalizing said second affinity to generate a second vote; and

- combining the first and second votes to obtain a score, wherein the score reflects the overall affinity of said candidate peptide for said target protein.

91. (New) The method of Claim 90, wherein said protein is a MHC class I protein and wherein said peptide comprises an epitope whereby the MHC class I protein binds to the peptide.

92. (New) The method of Claim 90, wherein said first and second affinity predictions are predicted by a method selected from the group comprising quadratic programming, linear programming, anchor scoring, and profile-based scoring, and wherein the second method is not the same method selected for the first method.

93. (New) A method of predicting the binding strength of a candidate peptide for a target protein, said method comprising:

- obtaining sequence data for a candidate peptide;

- obtaining sequence and binding strength data for at least one peptide of known affinity for a target protein;

generating a first vote by scaling a first affinity prediction of the candidate peptide for said target protein;

generating a second vote by scaling a second affinity prediction of the candidate peptide for said target protein, wherein at least one of said first and said second predictions uses the sequence and binding strength data for the at least one peptide of known affinity for said target protein; and

combining the first and second votes to create a score, wherein said score reflects the relative binding strength of the candidate peptide for said target protein.

94. (New) The method of Claim 93 further comprising:

ordering the candidate peptide so as to create a list according to the candidate peptide's score, wherein a candidate peptide with a largest score is first in said list and a candidate peptide with a lowest score is last in said list;

separating said list into a R1, a R2, and a R3 rankings, wherein said R1 ranking is defined as a point in the list where a number of moderate binding strength peptides is greater than a number of high binding strength peptides, wherein said R2 ranking is defined as a point in the list where a number of low binding strength peptides is greater than the sum of a number of moderate and high binding strength peptides, and wherein said R3 ranking is defined as a point in the list for a last low binding strength peptide;

assigning a binding level class of high, moderate, low, or none to each candidate peptide on the list based on if it is above R1, between R1 and R2, between R2 and R3, and below R4 respectively; and

selecting a candidate peptide from the list based on the assigned binding level class associated with the candidate peptide, thereby selecting a peptide with a desired level of affinity for a target protein.

95. (New) The method of Claim 94, wherein said target protein is a MHC class I protein and wherein said peptide comprises an epitope whereby said MHC class I protein binds to said peptide.

96. (New) The method of Claim 94, wherein generating the vote from the affinity predictions comprises a method selected from the group consisting of 1) linearly scaling each affinity prediction so that the vote has a value between 1 and 0, 2) nonlinearly scaling each affinity prediction so that the vote has a value between 1 and 0, and 3) scaling each affinity

prediction in a manner so a particular type of method can have a different weight in voting, wherein the value is maintained between 1 and 0.

97. **(New)** The method of Claim 94, wherein said first and second affinity predictions are predicted by a method selected from the group comprising quadratic programming, linear programming, anchor scoring, and profile-based scoring, and wherein the second method is not the same method selected for the first method.

98. **(New)** The method of Claim 94, wherein the high binding strength peptide is defined as one with an  $IC_{50}$  of less than 1nM, wherein the moderate binding strength peptide is defined as one with an  $IC_{50}$  of more than 1nM and less than 100nM, wherein the low binding strength peptide is defined as one with an  $IC_{50}$  of more than 100nM and less than 10  $\mu$ M, and wherein no binding is defined as an  $IC_{50}$  of more than 10  $\mu$ M.

99. **(New)** The method of Claim 94, wherein the method is repeated for a second candidate peptide, thereby generating a second score for the second candidate peptide, wherein said score is used to order the second candidate in the list.

100. **(New)** The method of Claim 94, wherein the binding strength data for at least one peptide of known affinity for a target protein is selected from the group comprising a dissociation constant,  $k_a$ ,  $k_d$ , or  $IC_{50}$ .

101. **(New)** The method of Claim 97, wherein said profile-based scoring uses a clustering heuristic selected from the group comprising iterative multiple alignment, letter frequencies, and position dependencies reflected by two (2) tests.

102. **(New)** The method of Claim 101, wherein said profile-based scoring employs a principle selected from the group comprising dimensionality reduction, multiple intra-allelic motifs, and anchor selection.

103. **(New)** A method for generating an epitope useful in the treatment of cancer, said method comprising:

- obtaining sequence and binding strength data for at least one epitope of known affinity for a MHC class I protein;

- obtaining sequence data for a candidate epitope for a MHC class I protein;

- predicting a first affinity for the candidate epitope with said target MHC class I protein using a first method;

predicting a second affinity for the candidate epitope with said target MHC class I protein using a second method, wherein at least one of said first and said second methods uses the data for the epitope of known affinity;

attributing a vote to each of said first and second affinities;

combining the two votes to obtain a vote total for the candidate epitope;

ordering the candidate epitope in a list according to the candidate epitope's vote total, wherein a candidate epitope with a highest vote total is first in said list and a candidate epitope with a lowest vote total is last in said list;

separating said list into a R1, a R2, and a R3 ranking, wherein said R1 ranking is defined as a point in the list where a number of moderate binding strength peptides is greater than a number of high binding strength peptides, wherein said R2 ranking is defined as a point in the list where a number of low binding strength peptides is greater than the sum of a number of moderate and high binding strength peptides, and wherein said R3 ranking is defined as a point in the list for a last low binding strength peptide, and wherein the high binding strength peptide is defined as one with an  $IC_{50}$  of less than 1nM, wherein the moderate binding strength peptide is defined as one with an  $IC_{50}$  of more than 1nM and less than 100nM, wherein the low binding strength peptide is defined as one with an  $IC_{50}$  of more than 100nM and less than 10  $\mu$ M, and wherein no binding is defined as an  $IC_{50}$  of more than 10  $\mu$ M;

assigning a binding level class of high, moderate, low, or none to each candidate epitope in the list based on if it is above R1, between R1 and R2, between R2 and R3, and below R4 respectively;

selecting the candidate epitope from the list based on the assigned binding level class associated with the candidate epitope, wherein said assigned binding level class is either in the moderate or lower levels of the binding classes; and

producing or isolating a protein that comprises said epitope, thereby generating an epitope that is useful in the treatment of cancer.

104. (New) An iterative multiple alignment (*aln*) method of predicting the relative binding affinity of a peptide for a MHC protein, comprising:

obtaining sequence and affinity information for a set of known epitopes for a MHC protein;

deriving a motif from said information for a set of known epitopes via an iterative multiple alignment heuristic, wherein said iterative multiple alignment heuristic builds a profile by adding a new sequence to an existing cluster until a stop condition is encountered;

generating a score for said peptide based on its similarity to said motif, wherein the iterative multiple alignment heuristic is also used on said peptide; and

predicting the relative binding affinity of said peptide for said MHC protein based on said score.

105. (New) A letter frequency (LetFq) method of predicting the relative binding affinity of a peptide for a MHC protein, comprising:

obtaining sequence and affinity information for a set of known epitopes for said MHC protein;

deriving a motif from said information for a set of known epitopes via a letter frequency heuristic, wherein said letter frequency heuristic recursively splits a sequence cluster into two disjoint subclusters, wherein said splitting is according to a sequence letter at a chosen profile position, and wherein said letter frequency heuristic repeats the previous procedure for the two subclusters thus formed;

generating a score for said peptide based on its similarity to said motif; and

predicting the relative binding affinity of said peptide for said MHC protein based on said score.

106. (New) A method based on  $\chi^2$  statistical significance ( $Ki^2$ ) tests for predicting the relative binding affinity of a peptide for a MHC protein, comprising:

obtaining sequence and affinity information for a set of known epitopes for said MHC protein;

deriving a motif from said information for a set of known epitopes according to dependencies between peptide positions, revealed by a  $Ki^2$  significance test, wherein said  $Ki^2$  significance test recursively splits a sequence cluster into two disjoint subclusters, wherein said splitting is according to a sequence letter at a chosen profile position, and wherein said  $Ki^2$  significance test repeats the previous procedure for the two subclusters thus formed;

generating a score for said peptide based on its similarity to said motif; and

predicting the relative binding affinity of said peptide for said MHC protein based on said score.

107. (New) A method for assessing the binding affinity between a candidate epitope and a MHC protein, said method comprising:

- obtaining a collection of sequence and binding strength data for at least one epitope of known affinity for said MHC protein;

- obtaining sequence data for said candidate epitope;

- predicting a first binding affinity for the candidate epitope for said MHC protein using a first predictive method, wherein said prediction is based, in part, upon the candidate epitope sequence data;

- predicting a second binding affinity for the candidate epitope for said MHC protein using a second predictive method, wherein said prediction is based, in part, upon the candidate peptide sequence data, said second predictive method further comprising the use of the collection of sequence and binding strength data for the at least one epitope of known affinity for said MHC protein, and wherein said second predictive method is different from said first predictive method;

- combining said first and second binding affinities; and

- evaluating the combined first and second binding affinities as a basis for assessing the binding affinity between the candidate epitope and the MHC protein.

108. (New) The method of Claim 107, wherein the MHC protein is a MHC class I protein.